

2.4. Incubation in Simulated Body Fluid (SBF)

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An abbreviated version of this protocol was published in Materials in Apr 2021
Inverse 3D Printing with Variations of the Strand Width of the Resulting Scaffolds for Bone Replacement
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Detailed protocol

Simulated body fluid (SBF) (500 ml) was prepared as described by Jalota et al [1]. The chemicals used to prepare the SBF and the amounts added are listed in Table 1. A beaker of deionized water was placed on a magnetic stirrer at 37°C. Each chemical was weighed using an electronic balance and added to the deionized water in the order shown in Table 1. An electronic pH meter (Mettler Toledo, EL20, Columbus, OH, USA) was then used to measure the exact pH of the solution, and hydrochloric acid was slowly added until the solution reached a pH of 7.4. The beaker was then covered with aluminum foil and left on the magnetic stirrer overnight. The next day, the solution was filtered through a 0.2 µm (pore size) filter and sterile sealed (ready for use).

Table 1: Composition of SBF (according to Jalota et al. [1])

Reagent	Obtained by	Art.No.	Quantity [g]
Sodium chloride (NaCl)	Sigma-Aldrich	S9888	3.274
Sodium hydrogen carbonate (NaHCO ₃)	Sigma-Aldrich	S6014	1.134
Potassium chloride (KCl)	Sigma-Aldrich	P3911	0.187
di-sodium hydrogen phosphate dihydrate (Na ₂ HPO ₄ ·2H ₂ O)	Sigma-Aldrich	71643	0.089
magnesium chloride hexahydrate (MgCl ₂ ·6H ₂ O)	Sigma-Aldrich	M8266	0.071
calcium chloride dihydrate (CaCl ₂ ·2H ₂ O)	Sigma-Aldrich	223506	0.184
sodium sulphate (Na ₂ SO ₄)	Sigma-Aldrich	239313	0.0355
Tris ((CH ₂ OH) ₃ CNH ₂)	Sigma-Aldrich	T1378	3.0285
1M HCl	Supelco	1090571000	Until pH 7.4

- Jalota, S.; Bhaduri, S.B.; Tas, A.C. Using a synthetic body fluid (sbf) solution of 27 mm hco₃⁻ to make bone substitutes more osteointegrative. *Materials Science and Engineering: C* **2008**, 28, 129-140. <http://dx.doi.org/10.1016/j.msec.2007.10.058>

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- Seidenstuecker, M., Schilling, P., Ritschl, L., Lange, S., Schmal, H., Bernstein, A. and Esslinger, S. (2021). Inverse 3D Printing with Variations of the Strand Width of the Resulting Scaffolds for Bone Replacement. *Materials* 14(8). DOI: [10.3390/ma14081964](https://doi.org/10.3390/ma14081964)

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